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Commentary: EPA's proposed expansion of dose-response analysis is a positive step towards improving its ecological risk assessment☆

Evgenios Agathokleous [a, b, *], Alessandro Anav [c], Valda Araminiene [d], Alessandra De Marco [e], Marisa Domingos [f], Mitsutoshi Kitao [a], Takayoshi Koike [b], William J. Manning [g], Elena Paoletti [c], Costas J. Saitanis [h], Pierre Sicard [i], Marcello Vitale [j], Wenjie Wang [k, l], Edward J. Calabrese [m]

[a] Hokkaido Research Center, Forestry and Forest Products Research Institute (FFPRI), Forest Research and Management Organization, 7 Hitsujigaoka, Sapporo, Hokkaido, 062-8516, Japan
[b] Research Faculty of Agriculture, Hokkaido University, Kita 9 Nishi 9, Sapporo, Hokkaido, 060-8589, Japan
[c] National Council of Research, Via Madonna del Piano 10, Sesto Fiorentino, Florence, 50019, Italy
[d] Institute of Forestry, Lithuanian Research Centre for Agriculture and Forestry, Girionys, 53101, Kaunas district, Lithuania
[e] Italian National Agency for New Technologies, Energy and the Environment (ENEA), C.R. Casaccia, S. Maria di Galeria, Rome, 00123, Italy
[f] Instituto de Botânica, Núcleo de Pesquisa em Ecologia, PO Box 68041, 04045-972, SP, Brazil
[g] Department of Plant, Soil and Insect Sciences, University of Massachusetts, Amherst, MA, USA
[h] Lab of Ecology and Environmental Science, Agricultural University of Athens, Iera Odos 75, Athens, 11855, Greece
[i] ARGANS, 260 route du Pin Montard, Sophia Antipolis cedex, 06904, France
[j] Department of Environmental Biology, Sapienza University of Rome, Piazzale Aldo Moro 5, Rome, 00185, Italy
[k] Northeast Institute of Geography and Agroecology, Chinese Academy of Sciences, Changchun, 130102, China
[l] Northeast Forestry University, Harbin, 150040, China
[m] Department of Environmental Health Sciences, Morrill I, N344, University of Massachusetts, Amherst, MA, 01003, USA

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ABSTRACT

The United States Environmental Protection Agency (US EPA) has recently proposed changes to strengthen the transparency of its pivotal regulatory science policy and procedures. In this context, the US EPA aims to enhance the transparency of dose-response data and models, proposing to consider for the first time non-linear biphasic dose-response models. While the proposed changes have the potential to lead to markedly improved ecological risk assessment compared to past and current approaches, we believe there remain open issues for improving the quality of ecological risk assessment, such as the consideration of adaptive, dynamic and interactive effects. Improved risk assessment including adaptive and dynamic non-linear models (beyond classic threshold models) can enhance the quality of regulatory decisions and the protection of ecological health. We suggest that other countries consider adopting a similar scientific-regulatory posture with respect to dose-response modeling via the inclusion of non-linear biphasic models, that incorporate the dynamic potential of biological systems to adapt (i.e., enhancing positive biological endpoints) or maladapt to low levels of stressor agents.

1. Introduction

While the history of dose-response assessment can be traced back to the late 19th century, its application for risk assessment began in the early 20th century with the development of public drinking water (US Public Health Service, 1925) and occupational health standards for chemicals and ionizing radiation (Calabrese, 2009).

During the mid-decades of the 20th century, there was a substantial expansion of recommended occupational health standards at the state level, and by national/international advisory groups. In 1970, the US Occupational Health and Safety Administration adopted several hundred recommended health standards of the American Conference of Governmental Industrial Hygienists,

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* Corresponding author. Hokkaido Research Center, Forestry and Forest Products Research Institute (FFPRI), Forest Research and Management Organization, 7 Hitsujigaoka, Sapporo, Hokkaido, 062-8516, Japan.
E-mail addresses: evgenios@affrc.go.jp, globalscience@frontier.hokudai.ac.jp (E. Agathokleous).

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essentially all following a threshold dose-response model. By the end of the 1970s, all regulatory agencies in the US had transitioned to two-tier approach: one, with threshold, for non-carcinogens, and another, linear no-threshold (LNT), for carcinogens (Calabrese, 2009, 2018a).

These regulatory actions initially assumed a threshold dose-response model. The threshold model was generally recognized and accepted over the first several decades of the 20th century. The threshold dose-response model concept would be first challenged following the report of Muller in 1927 that X-rays produced gene mutations in fruit flies (Muller, 1927). In 1930, Muller proposed the concept of a Proportionality Rule (i.e. linear dose-response) to characterize the effects of ionizing radiation on assumed gene mutation, based on transgenerational phenotypic changes in fruit flies (Muller, 1930). Over the next 25 years there was a growing tension, amongst some members of the radiation genetics and medical communities over whether the threshold dose-response model should be replaced by a linear one for radiation (and later chemicals) inducing gene mutation (Calabrese, 2016, 2018b). After numerous disputes, a major change occurred in 1956 when the US National Academy of Sciences Biological Effects of Atomic Radiation (BEAR-1) Genetics Panel declared in an influential and well-publicized report that there was no threshold for assumed gene mutations in reproductive cells (Calabrese, 2017a, 2017b, 2018a). The idea promoted by BEAR-1 Genetics Panel was generalized to somatic cells and applied to cancer risk assessment, within several years (Calabrese, 2014). These changes led to a two tier system for risk assessment, one for carcinogens following a linear model and one for non-carcinogens following a threshold model. The linear model assumed that induced gene mutations were not repairable and the damage was cumulative, leading to the belief that total dose, rather than dose rate, was the best predictor of gene mutation. The LNT model was adopted by the US Environmental Protection Agency (EPA) for carcinogen risk assessment for ionizing radiation and chemical carcinogen in the mid- 1970s, and continues to the present.

With respect to ecological risk assessment (ERA) (Supplementary Materials, Fig. 1S), the issue was not recognized as significant as in the case of cancer and human toxicology. As a result of this differential viewing of human versus ecological effects, the general course of action for ERA was framed by the assumption of the threshold dose-response model, during the later decades of the 20th century (Calabrese and Baldwin, 1993). However, over this time period new perspectives and challenges emerged that could affect the process of carcinogen and non-carcinogen risk assessment for humans, and for ecological receptors. A potential significant development involved the rediscovery and substantial documentation of the hormetic-biphasic dose-response that may be applied to human health and to ERA for chemicals and radiation (Calabrese, 2001, Calabrese and Baldwin, 2000a,b, 2001; Vom Saal, 2007; Calabrese 2008, 2011; see www.BELLEonline.com (newsletters) for a substantial number of scholarly articles on this topic both supporting and challenging the approaches of hormesis in risk assessment). A second significant development was the concept that linear dose responses (Calabrese, 2017a, 2018a) may be applied to non-cancer endpoints (Bogen, 2016, 2017) such as lead-induced neurotoxicity (USEPA, 1998), particulate matter effects on humans (WHO, 2006; USEPA, 2013; Enstrom, 2017), effects of endocrine disruptor agents on humans (Zoeller et al., 2015; Schug et al., 2016), and ozone-induced plant injury (Agathokleous et al., 2019a).

To date, major regulatory agencies have adopted the LNT dose-response model for the particulate matter effects on humans (WHO, 2006; USEPA, 2013), and for the ozone effects on plants to derive critical levels for vegetation protection (Agathokleous et al., 2019a). The adoption of the LNT for these two particular agents is significant by itself, but also important because it can trigger such adoptions by other regulatory changes concerning other agents and endpoints.

2. Analysis

The nature of the dose-response relationship is a critical factor guiding the risk assessment and eventually driving the estimates of potential risk. While risk assessment has long been based upon linear or threshold dose-response relationships, it is now recognized that linear and threshold dose-response relationships are often not the norm and that the dose-response relationship can be non-linear polyphasic (Costantini et al., 2010; Calabrese, 2017c; Calabrese and Mattson, 2017; Agathokleous et al., 2018a, 2019a,b; Kim et al., 2018; Leak et al., 2018) (note: for the debate on hormesis, the reader may also refer e.g. to Kefford et al., 2008; Ricci and Sammis, 2012; Sacks and Siegel, 2016, 2017; Mushak, 2013a,b; Jargin, 2018; www.BELLEonline.com - see newsletters). In light of the significance of these advances in regulatory science, the EPA has recently proposed changes to strengthen the transparency of its regulatory science[1] (USEPA, 2018). Of relevance to the present paper is that the EPA proposes moving away from the use of linearity as a default model for risk assessment, providing for the first time an opportunity to consider non-linear functions beyond the threshold model.[2]

Current scientific findings indicate that non-linear dose response modeling is more biologically plausible than low dose linear modeling (Costantini et al., 2010; Calabrese and Mattson, 2017; Agathokleous et al., 2018a, 2019a; Kim et al., 2018; Leak et al., 2018). Hence, we believe that consideration of alternative models has the potential to enhance the quality of both risk estimates and subsequent risk management decisions that guide the derivation of environmental standards and cost-benefit assessments.

In the last two decades it was extensively shown that biphasic (e.g. *U*-shaped, *J*-shaped, and bell-shaped) dose responses are induced by a variety of chemical agents and environmental factors in a wide spectrum of endpoints and biological models, not only at individual but also at community level, including key ecological endpoints such as biodiversity, soil respiration, emission of leaf volatile organic compounds (VOCs), gas fluxes from soils, demography of pests of agricultural crops, and reproduction of terrestrial and aquatic organisms (Costantini et al., 2010; Schreck, 2010; Calabrese and Blain, 2011; Calabrese and Mattson, 2017; Agathokleous, 2018; Agathokleous et al., 2018a,b, 2019a; Kim et al., 2018; Leak et al., 2018; Sial et al., 2018; Yue et al., 2018). We thus believe consideration of non-linear models has the potential to significantly improve risk estimation.[3]

[1] The first part of the EPA proposal excludes scientific studies whose data are not publicly accessible. Many scientific societies have not supported the EPA proposal, and thousands of comments have been submitted criticizing the study exclusion part (EPA, Docket ID: EPA-HQ-OA-2018-0259, https://www.regulations.gov/docket?D=EPA-HQ-OA-2018-0259). This paper is not intended to appraise the study exclusion part of the EPA proposal.

[2] EPA mentions "EPA should also incorporate the concept of model uncertainty when needed as a default to optimize low dose risk estimation based on major competing models, including linear, threshold, and U-shaped, J-shaped, and bell-shaped models" (USEPA, 2018). While it is unclear what "when needed" is supposed to mean, we suggest that "when needed" is when the data are inadequate for confident modeling as in the case of substantial extrapolation beyond the observable data. For argumentation purposes, this would include extrapolation approximately two-fold beyond the observable data.

[3] For an alternative view on contested subjects, the reader may refer to the comments by Finkel (2018) on the EPA website.

The EPA states it should give "appropriate consideration"[4] to high quality studies that explore: i) "a broad class of parametric concentration-response models with a robust set of potential confounding variables"; ii) "nonparametric models that incorporate fewer assumptions"; iii) "various threshold models across the exposure range"; and iv) "spatial heterogeneity" (USEPA, 2018).

We consider this proposal an important step towards a more dynamic and flexible risk assessment process which will not view animals, plants, and ecosystems as static but as dynamic biological entities displaying complex responses to different environmental conditions. For instance, the need for dynamic risk assessment is highlighted for vegetation response to gases which is driven by soil water and nutrient availability (Körner, 2006; Proietti et al., 2016; Anav et al., 2018) as well as by meteorological seasonality, as it occurs in subtropical regions exposed to well defined wet and dry periods (Moura et al., 2014). In addition, two aspects may be considered in subtropical regions: i) pioneer tree species from forest plots may be more acclimated to the stressful environment than non-pioneer trees, presenting better conditions to perpetuate in the disturbed forest remnants; and ii) multilinear regression analyses revealed that the oscillations in the physiological leaf traits in both functional groups of tree species coincide with oscillations in both meteorological variables and air pollutants, notably air temperature, global solar radiation, ozone and nitrogen dioxide (Esposito et al., 2018). Interaction between variables (De Marco et al., 2014; Vitale et al., 2014) is an important reason for considering more complex dose-response functions and dynamic risk assessment.

With regard to proposition iii of the EPA, we believe that biological plausibility linked with appropriate experimental studies of high quality should guide low dose modeling strategies. This approach can enhance the likelihood of improved biodiversity risk estimates (Pereira et al., 2010). We therefore believe the EPA proposal is a positive step towards scientifically-based biologically motivated ERA. However, while this is a significant advance, we believe there remain open issues that need to be addressed in order to improve ERA:

- **Integration of adaptive response in ERA modeling:** Adaptive responses are a fundamental feature of evolution and these processes are often described by hormetic dose responses with specific quantitative features that are highly generalizable across aquatic and terrestrial organisms (Laughlin et al., 1981; Cedergreen et al., 2007; Costantini et al., 2010; Schreck, 2010; Calabrese and Blain, 2011; Hook et al., 2014; Ng et al., 2016; Calabrese and Mattson, 2017; Iavicoli et al., 2018; Agathokleous, 2018; Agathokleous et al., 2018a, 2019a,b; Oliveira et al., 2018; Leak et al., 2018). These findings support the use of hormesis and other non-linear dose-response models in terrestrial and aquatic ERA.
- **Integration of space/time variables in ERA modeling:** With respect to biphasic dose-response relationships, both space and time should be considered. Low-dose effects often show temporal fluctuations, with declining stimulatory responses over time (Agathokleous et al., 2019b; Leak et al., 2018; Sun et al., 2018).
- **Life histories of species role in ERA:** Low dose effects may be influenced by the life histories of species or ecotypes (McCallum et al., 2019). The developmental/ontogenic stage for which the dose-response is studied should be considered. For instance, this would mean that a potential net benefit should be considered at the long term, possibly after exposure throughout the biological cycle, or over several growing seasons for perennial plants. Therefore, a future target would be to incorporate the life history of the communities/populations for which the low-dose risk is estimated.
- **The endpoint matters:** Ecological systems are complex and a single endpoint may fail to reflect the most relevant outcome in ecological terms (Hook et al., 2014; Agathokleous et al., 2019a,c). By integrating multiple relevant endpoints, the capacity for classifying the low-dose effect as positive (beneficial) or negative (potentially harmful in the long run) may be enhanced (Hook et al., 2014; Agathokleous et al., 2019c).
- **Transgenerational ERA:** The present is not likely to mirror the future. Recent advances have revealed that effects of low doses on animals can affect the next generations via epigenetic mechanisms (Emborski and Mikheyev, 2018; Kishimoto et al., 2018; Mothersill and Seymour, 2018). This transgenerational preconditioning mode may lead to offspring with altered tolerance (increase or decrease) to stress. Therefore, risk estimates of one generation may not necessarily reflect risk estimates of next generations.
- **Interindividual variation:** The current ERA is based on experiments with controlled conditions and commonly where the subject organisms are at a healthy biological state. This raises important considerations whether the dose-response outcomes can be translated to real-world conditions. In fact, recent epidemiological studies have shown little or no evidence of ozone-induced injury in perennial vegetation (Agathokleous et al., 2019a). Furthermore, extensive research indicates that plant response to gases differs between open systems, where plants fully interact with the environment, and semi-closed systems, where the infrastructure creates and maintains a microenvironment different from the ambient conditions (Valkama et al., 2007; Feng et al., 2018). Notably, data from semi-closed systems comprised the base for present ERA. These examples suggest that environmental risk assessment can be improved with real-world or near real-world study conditions (Sicard et al., 2016).
- **Community level data needed in ERA:** Dose-response experiments at community level are scarce. The current risk assessment is principally based upon experiments where the response of individual organisms is studied. However, individual organisms may respond differently when coexisting with other individuals, so called forming populations (González-Fernández et al., 2017). A future target of risk assessment would be the implementation of dose responses where organisms are under the influence of competition with coexisting organisms, as typically happens in the nature.
- **Mixture toxicity and ERA:** Dose response modeling needs to address the issue of complex chemical mixtures (Belz et al., 2008; Ge et al., 2011; Zou et al., 2013; Mayo et al., 2015; Belz and Piepho, 2017; Docea et al., 2018; Liu et al., 2018).

3. Conclusions

This paper strongly supports the EPA proposal to move away from the use of LNT as a default in risk assessment and to consider non-linear models such as hormesis. It is scientifically necessary for EPA to incorporate evolutionarily based biologically motivated models for ecosystem risk assessment. Other countries should consider following this part of EPA's proposal.

Conflicts of interest

The authors declare no conflict of interest.

[4] It has not been properly defined by the EPA what "appropriate consideration" is supposed to mean. We consider "high quality studies" studies with robust experimental design, documented quality control, and with correct statistical testing and inference, with raw data being publicly available or not (see footnote 1).

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.envpol.2018.12.046.

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